

MeBOP

*Building Skills.*

*Bridging Barriers.*



- Khalid AB. Khumage
- MBBCh.
- Medical Resident at Zawia Teaching Hospital
- Zawia -Libya

# Zawia City





# *Scientific goals*

- Pursue a speciality training in internal medicine .
- Interested in : cardiology , hematology, oncology and infectious diseases.

# Scientific goals

- Importance of molecular biology in different clinical fields
- biological techniques have improved our understanding of the pathophysiology of various diseases and hence proper therapeutic options were made achievable

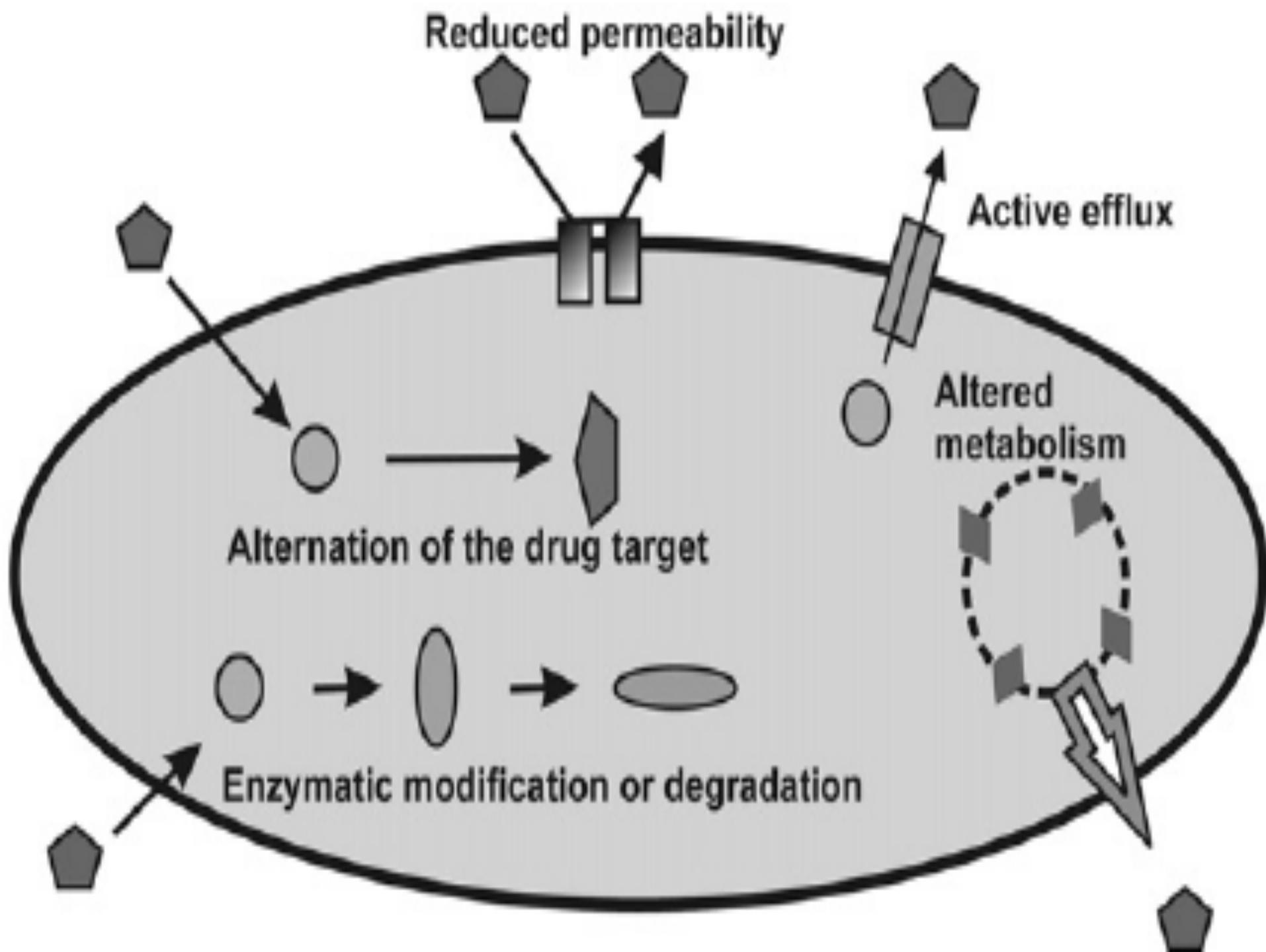
- Techniques ranging from DNA amplification (PCR) , recombenant DNA technology , gene analysis and cloning have revolutionized the biomedical sector and provided scientists with invaluable tools for research.

# Personal research interests

- As an internist , It is not uncommon to encounter cases of treatment failure in patients with infectious diseases, ICU patients and immunocompromised individuals .
- These cases can be challenging and demand a greater degree of collaboration between the physician , microbiologist and infectious disease specialist

# Personal research interests

- Understanding the different mechanisms of drug resistance in our local hospitals and communities is one of key research areas that I am concerned about
- Unfortunately , this kind of research usually requires resources that are beyond the local laboratorie`s potentials



# Biomedical research in Libya

- National center for medical research
- National center for disease control
- National Oncology Center



# Zawia Teaching Hospital

## مستشفى الزاوية التعليمي

Z.H

*zawia.hospital*



Z.H

# National Center for Disease control

- Is the official center concerned with research and studies covering both epidemic and endemic diseases , communicable and non- communicable diseases in Libya

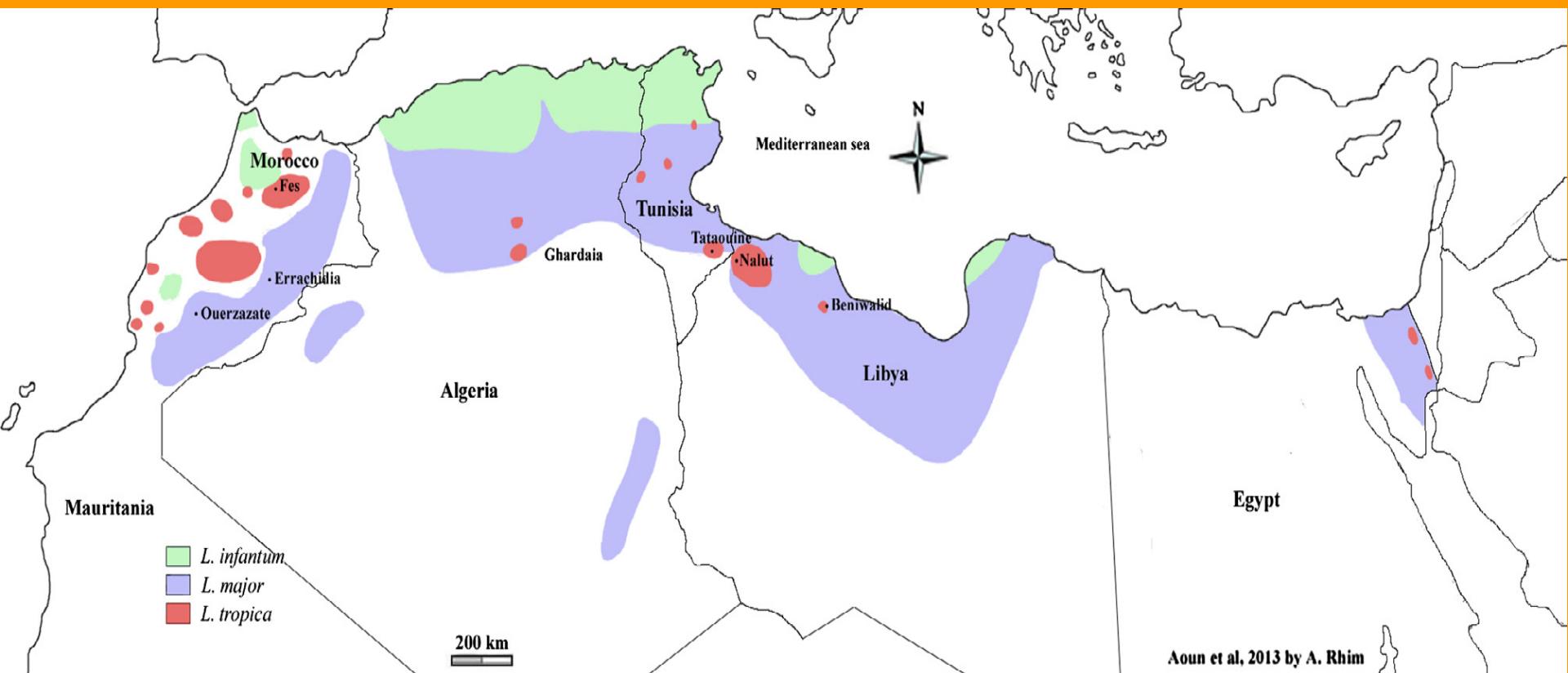
# National Center for Disease Control



# National Center for Disease Control

- With regards to parasitic diseases and aside from the common diarrheal infestations , two are considered as a major health problems in Libya ;
- 1) Cutaneus Leishmaniasis ( CL)
- 2) Echinococcosis ( hydatid disease)

# Leishmaniasis





# Diagnosis

- 1. Clinical Diagnosis: signs & symptoms  
Patient history (travel, vectors)

## 2. Laboratory Diagnosis :

# **Laboratory Diagnosis of leishmaniasis :**

## **Cutaneous leishmaniasis :**

- Tissue sample (scraping, aspirate or punch biopsy) for smear and culture

## **Visceral leishmaniasis :**

- Bone marrow biopsy or splenic aspirate for smear and culture.(N.N.N) V.L.(anemia , leukopenia , glubuline/albumine is high (Hypergammaglobulinia)
- Serology ( ELISA ) ( IFAT ).
- PCR
- Skin test
- Inoculate serum of infected person in lab. animals.

# *Animal inoculation*



Inoculate serum of infected person in lab. animals.



# Leishmaniasis

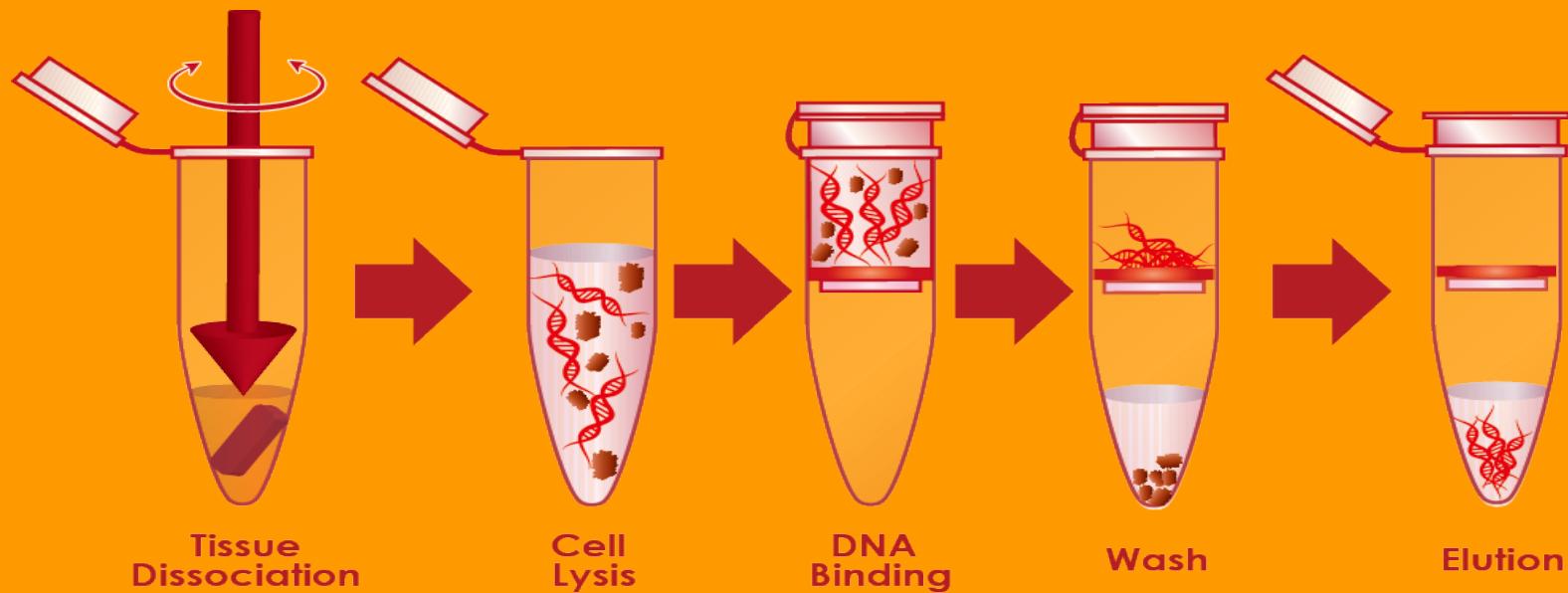
- In Libya , CL is widespread in the north-western region ( aljabal algharbi mountainous area ) . The first case was reported in 1930.
- The diagnosis of CL in Libya is based on clinical signs of the disease and microscopic observation of parasites in stained skin biopsies

# Leishmaniasis

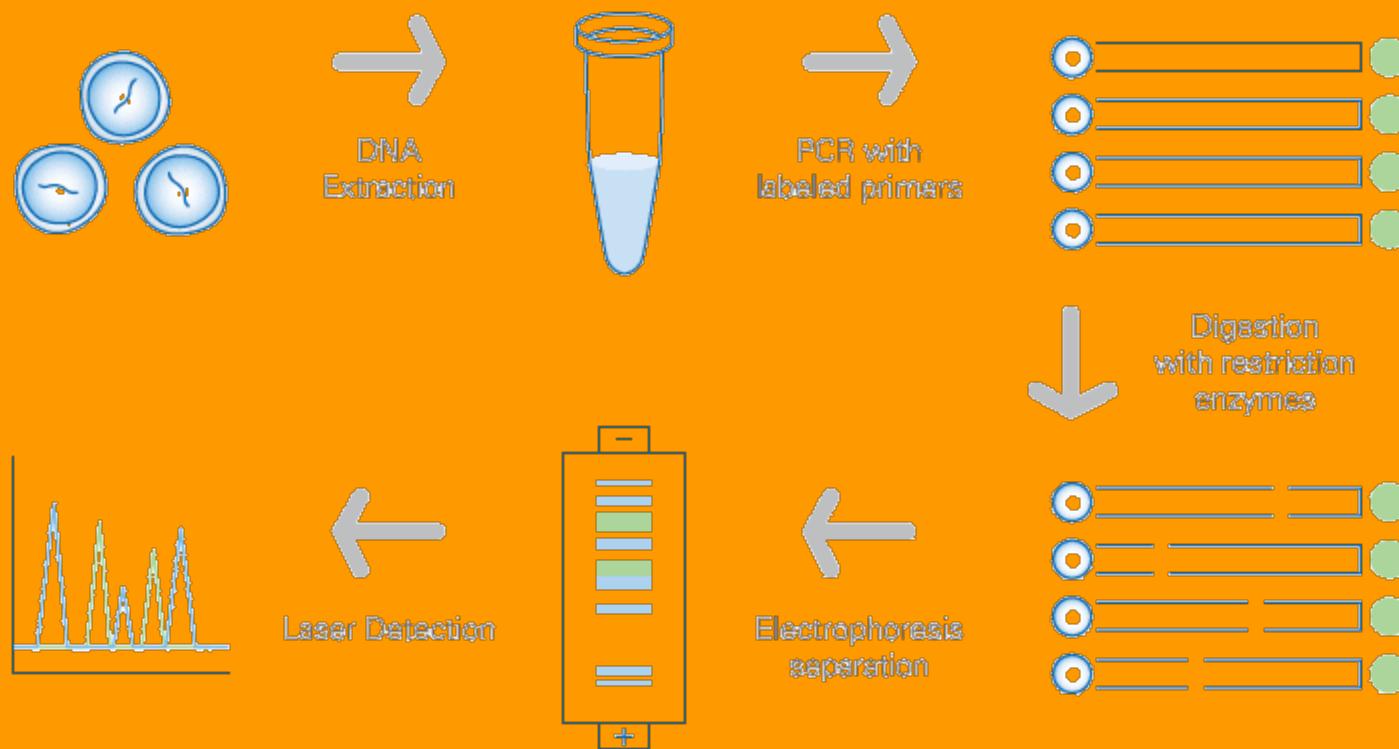
Specific and sensitive molecular diagnostic tools have not yet been implemented on a national level .

One study published by Amro A. et al in 2012 “ First Molecular Epidemiological Study of Cutaneus Leishmaniasis in Libya” has investigated the molecular characters of the parasite and its mode of transmission

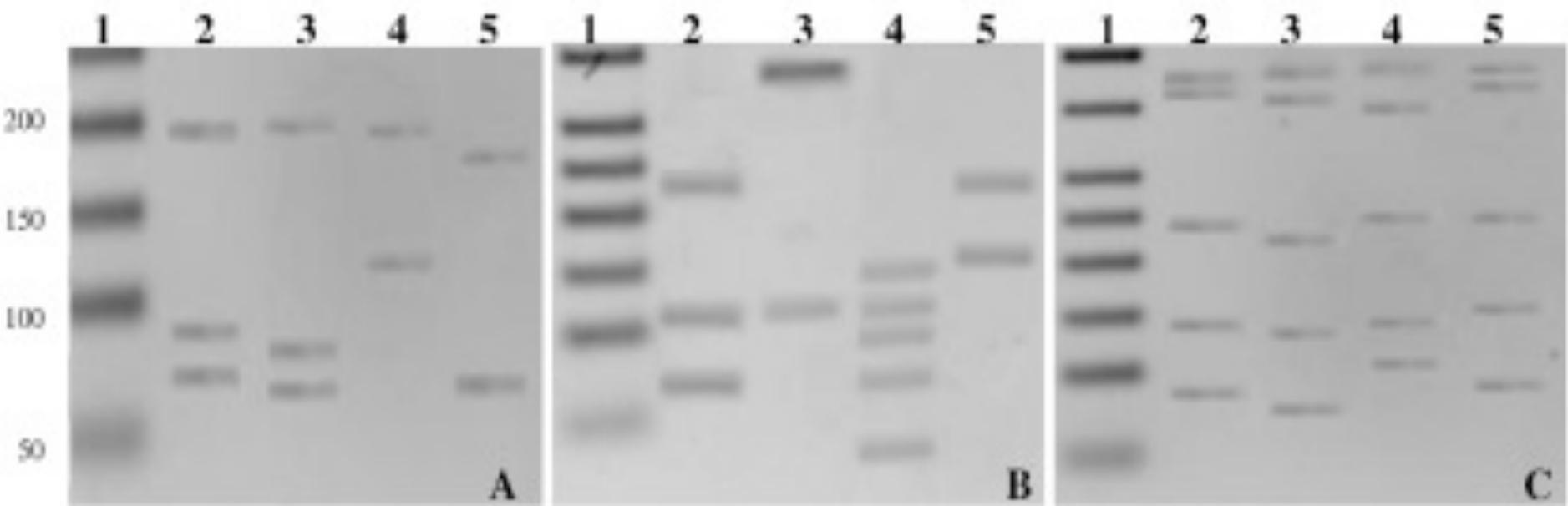
# DNA Extraction



# PCR-RFLP Approach



**A**; ITS1 PCR-RFLP profiles of different *Leishmania* species after digestion of products with HaeIII. **B**; ME PCR-RFLP profiles of different *Leishmania* species after digestion of EaeI. **C**; HSP70-PCR-RFLP profiles of different *Leishmania* species after digestion of products with HaeIII. 1; 50 bp molecular weight marker, 2; *L. donovani*, 3; *L. infantum*, 4; *L. major*, 5; *L. tropica* in all figures.

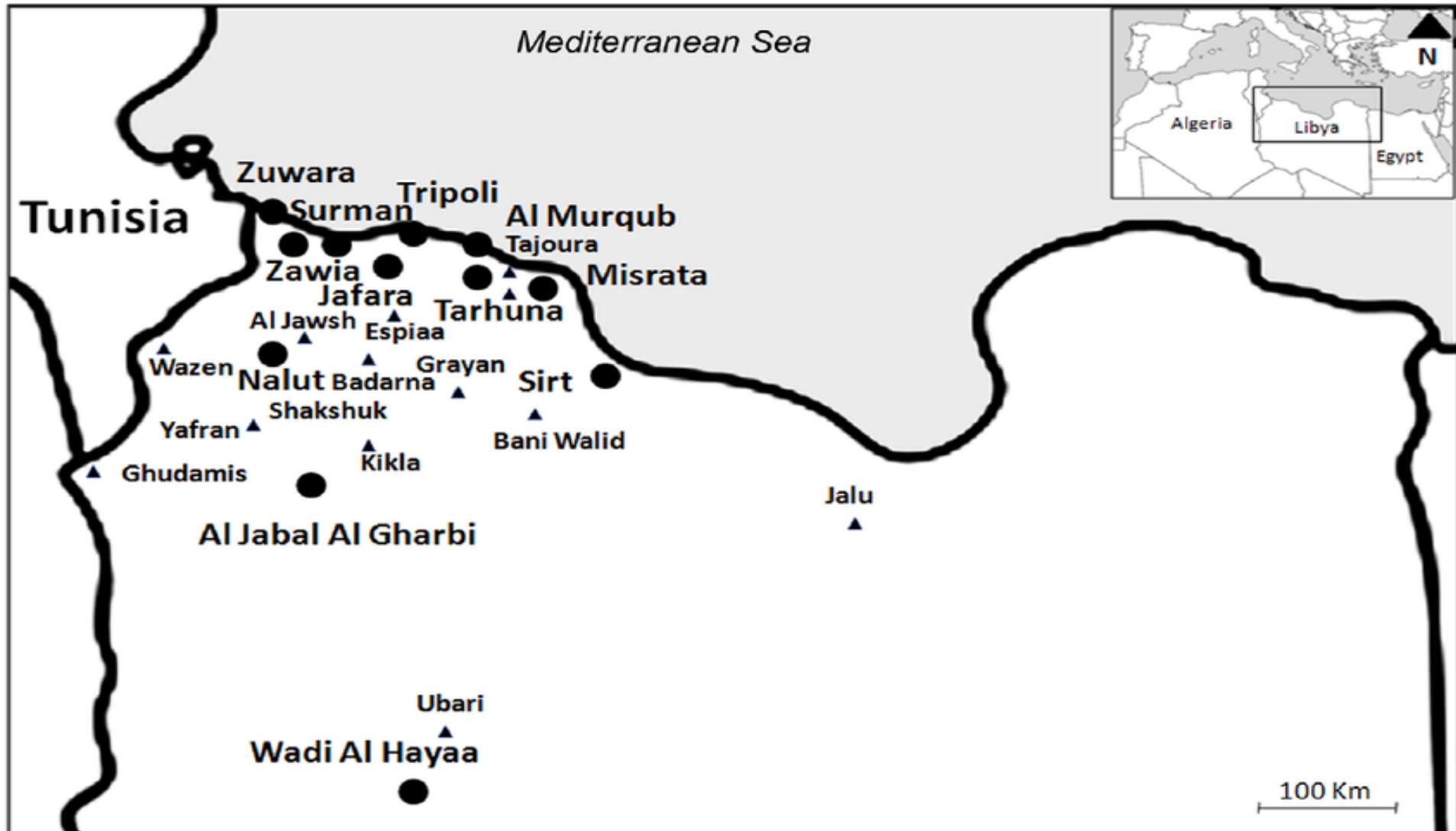


RFLP analysis of amplified internal transcribed spacer 1 ITS1 digested with restriction enzyme HaeIII and analysed by electrophoresis on agarose gel

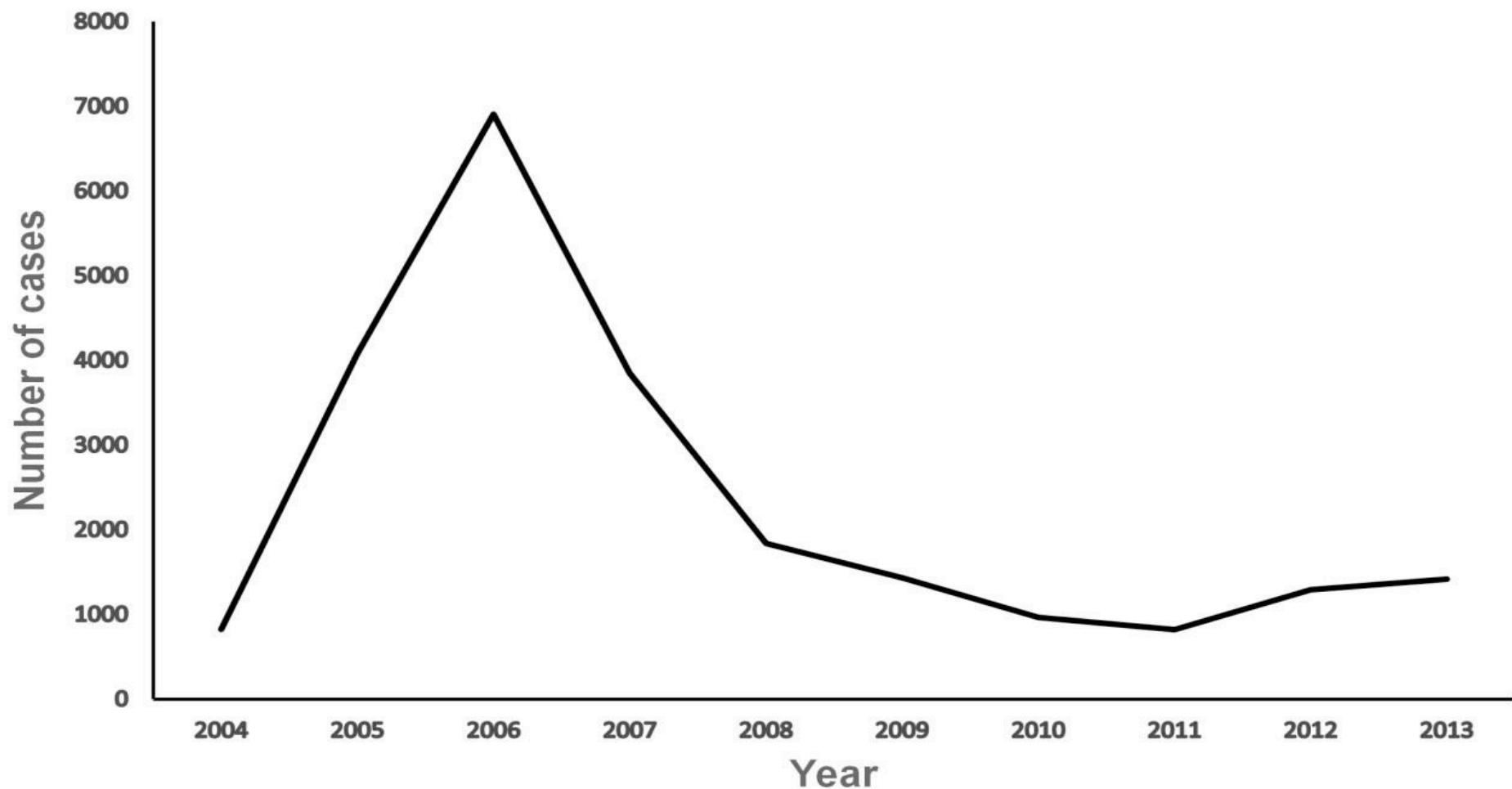
# Results

- The PCR – RFLP approach revealed that there are 2 distinct species of leishmania responsible for CL cases in Libya ;
- *Leishmanai major*
- *Leishmania tropica*

# Geographical distribution of CL in Libya



# Total annual number of cases reported to the Libyan National Centre for Disease Control 2004-2013



- In response to increasing number of CL cases during the last decades in Libya, a National Control Program was launched in 2006 which aims to prevent the eruption of epidemics Methods to control rodents were implemented and a wide scale campaign of vector control was also applied utilizing fogging and residual spraying of pyrethroids. Hence the number of cases has progressively decreased from 7180 in 2006 to 1800 in 2008 (LNCIDC reports).

# Current Situation

- The armed conflict which occurred in 2011 has affected all aspects of the life in the country. In Libya, the primary health care services were interrupted or massively impaired in some of the endemic areas. This has resulted in complete interruption of the national control program (NCP) in Libya.

# Current Situation

- Massive human migration from cities to villages and camps took place from May to October, the main CL transmission season. This is an additional risk factor because people were more prone to get bitten and infected . The quantification and the containment of these risk factors are major challenges .

# Haemoflagellates

## Leishmaniasis



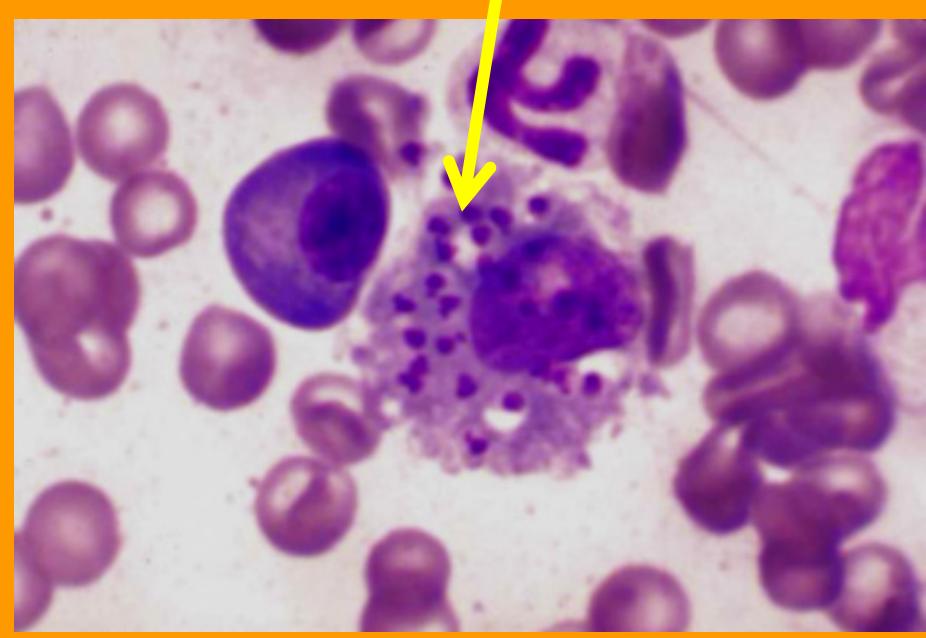
Leishmaniasis is a **zoonosis**.

Transmitted among mammalian hosts by female **sand flies**.



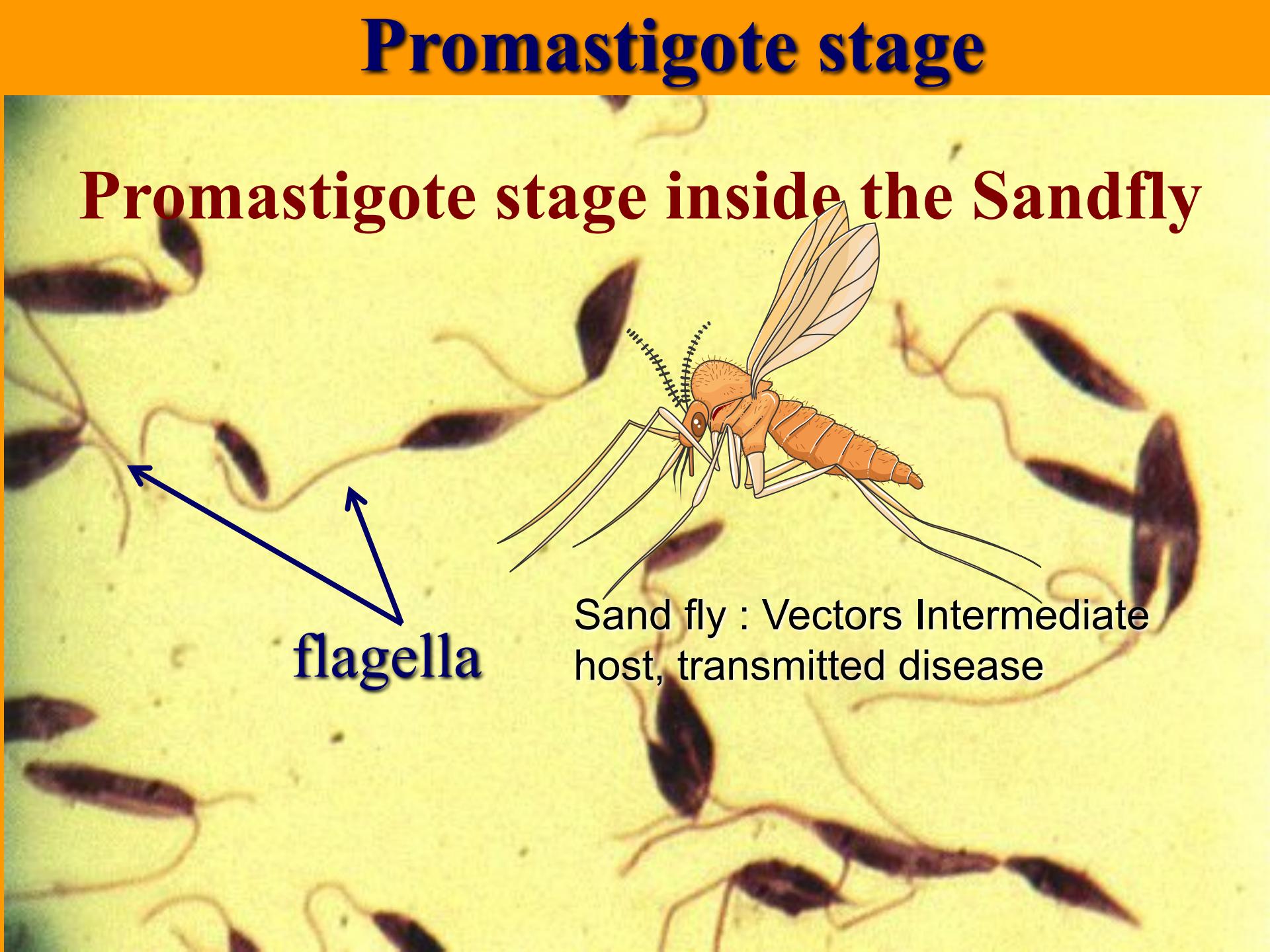
# Life Cycle of leishmaniasis

**Promastigote**  **Amastigote**  
*Transformation*

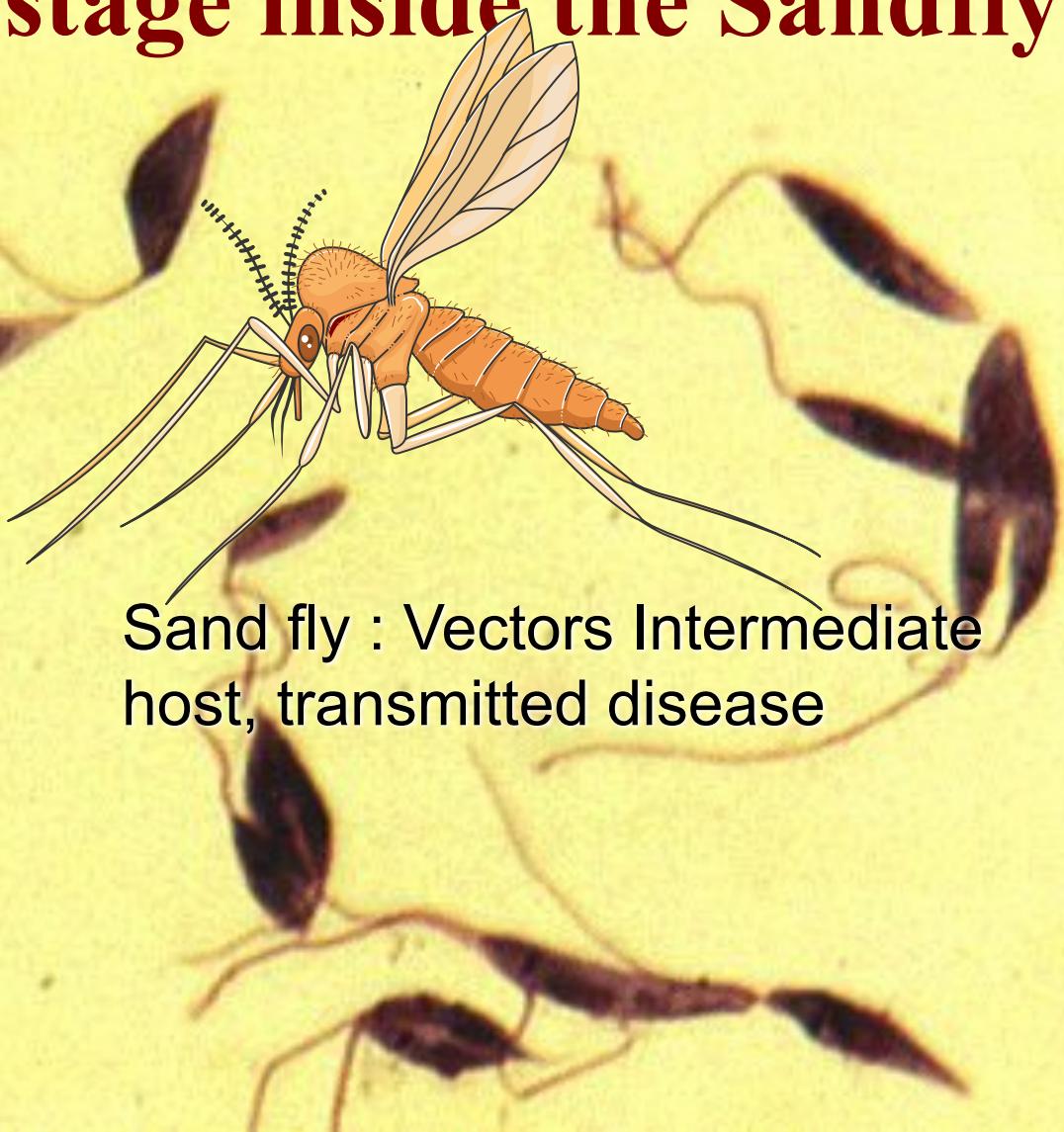


# Promastigote stage

Promastigote stage inside the Sandfly



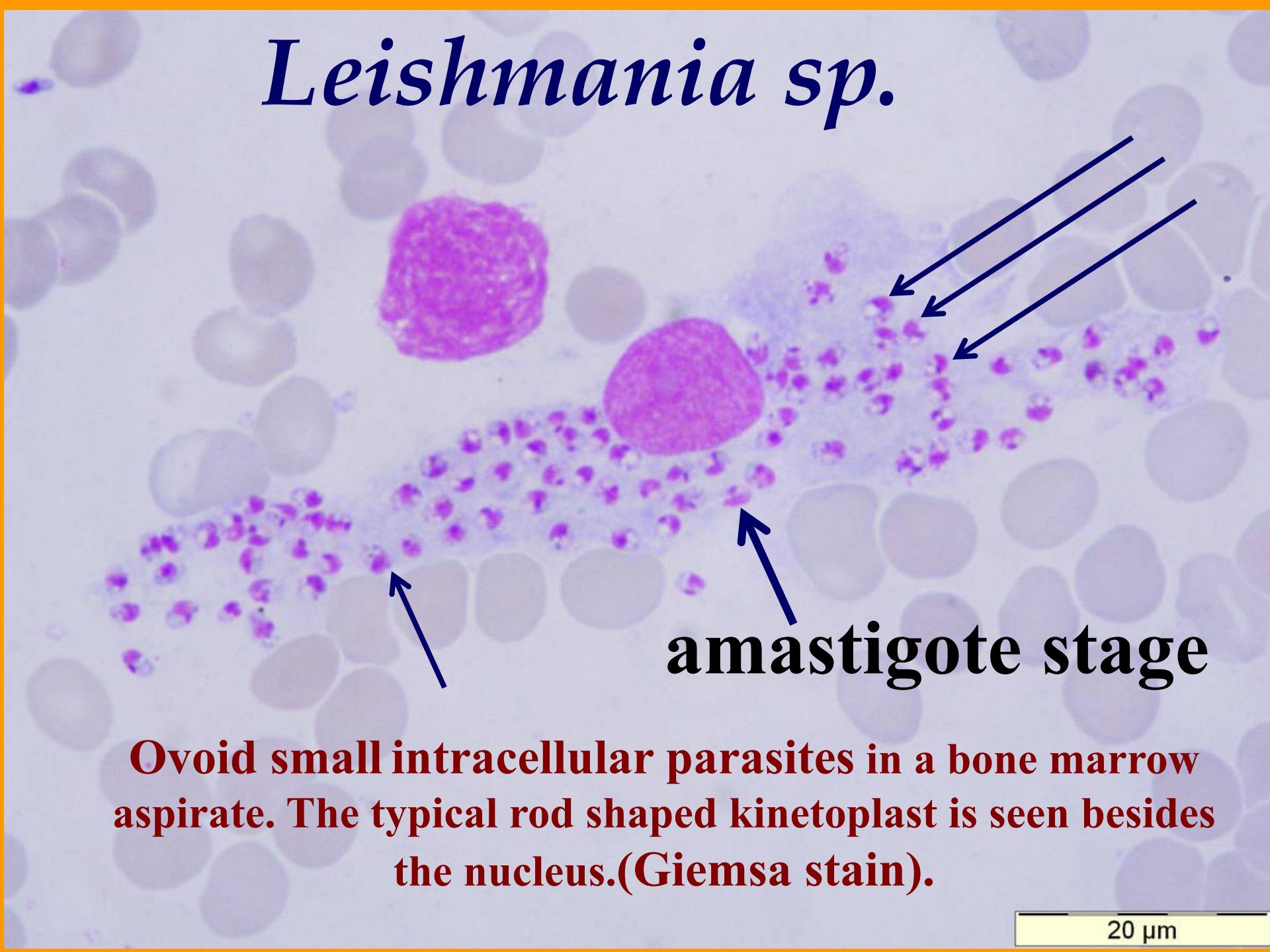
flagella



*Promastigotes in  
rosettes in a culture of  
an orient sore on N.N.N.  
medium (Giemsa stain).*



# *Leishmania* sp.



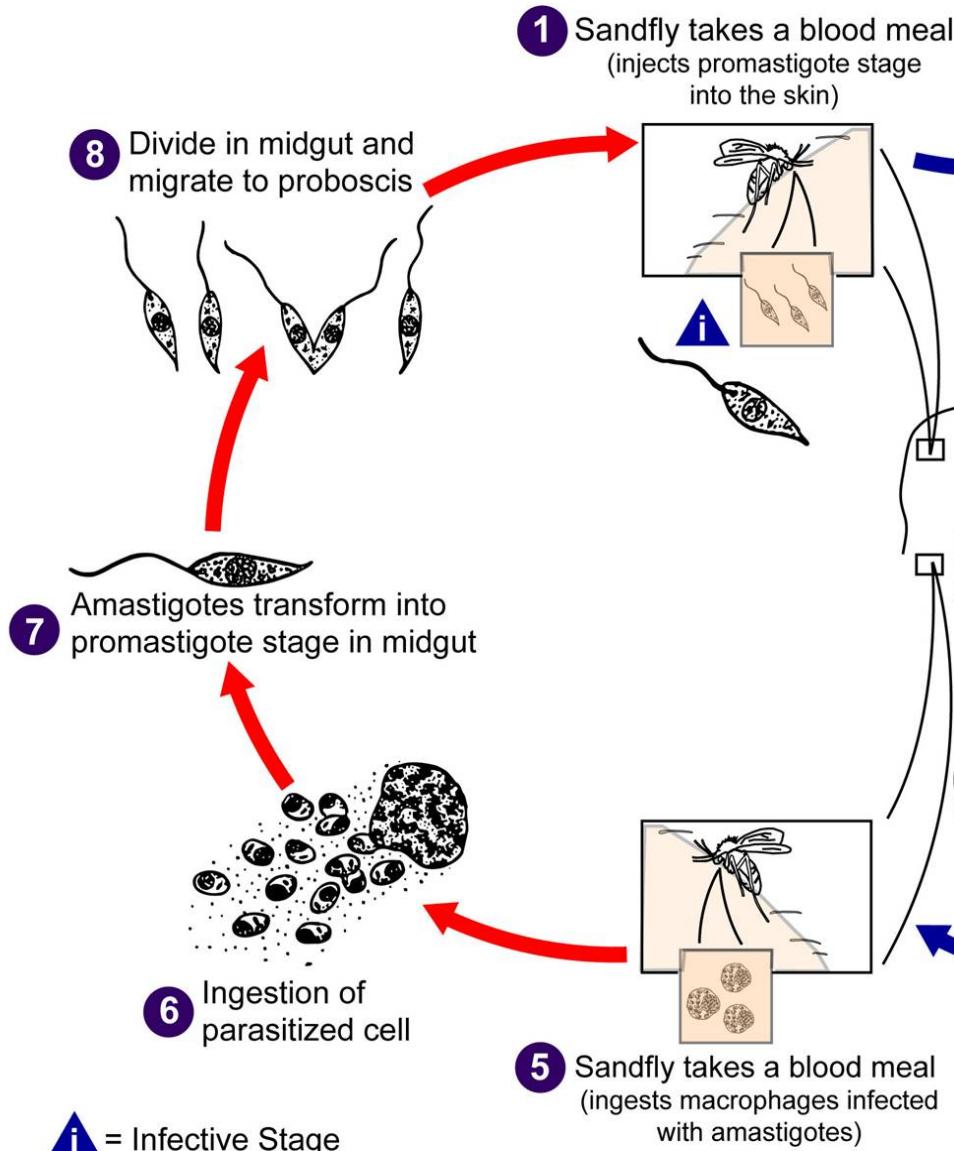
**amastigote stage**

**Ovoid small intracellular parasites in a bone marrow aspirate. The typical rod shaped kinetoplast is seen besides the nucleus.(Giemsa stain).**

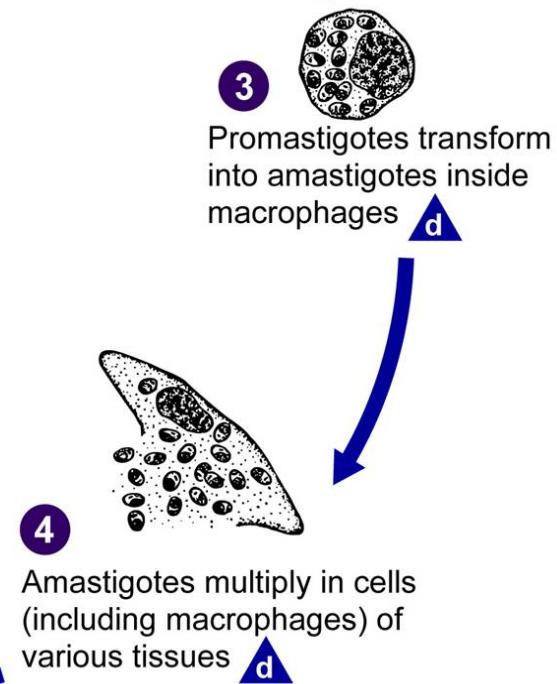
# Leishmaniasis

(*Leishmania spp.*)

## Sandfly Stages



## Human Stages



**i** = Infective Stage

**d** = Diagnostic Stage



anwendungsbereiches, ggf. mit ...

# Disease

# Cutaneous Leishmaniasis

**Cutaneous** forms of the disease normally produce skin ulcers on the exposed parts of the body such as the face, arms and legs. The disease can produce a large number of lesions



# A cutaneous leishmaniasis lesion on the arm.

**cutaneous leishmaniasis**



(by Mike Belosevic)

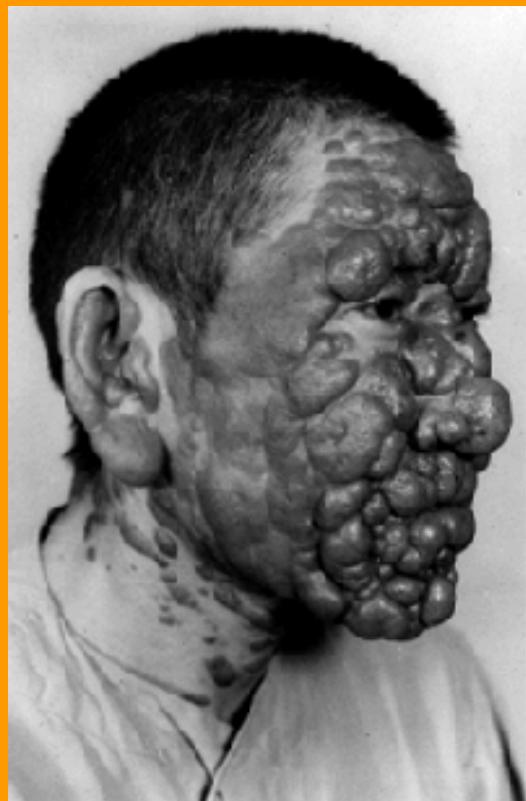
Some people have swollen lymph glands near the sores.

For example, the glands under the arm can swell if the sores are on the arm or hand.



The skin sores will **heal by themselves**, but this can take months or years. The sores can leave ugly scars.

# Cutaneous Leishmaniasis



# The Baghdad boil

## Baghdad-boil, 2004

Several hundred US soldiers  
in Iraq.



# *Leishmania tropica*



- Causes ulceration of the skin called Cutaneous Leshmaniasis
- Dry or urban C.L.
- Dry sore that may persist for several months before healing, then person is immune
- Some people “vaccinate” their children against Leshmaniasis.
- Rarely can cause infections of the viscera

# Mucocutaneous Leishmaniasis

Mucocutaneous leishmaniasis  
(Espundia)

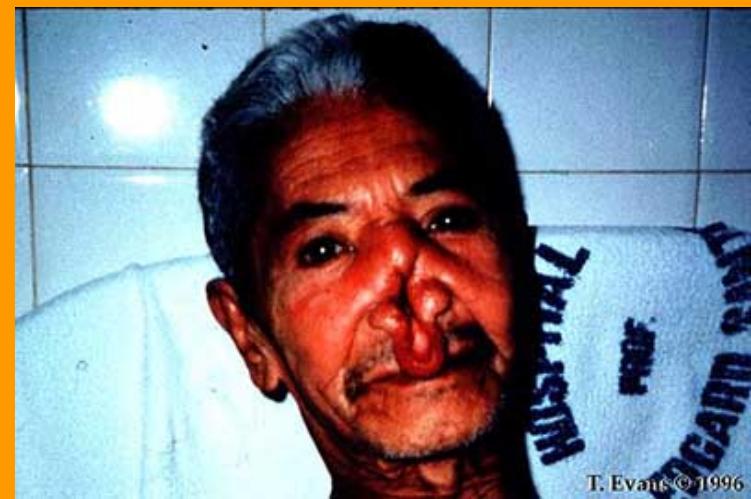
*Leishmania braziliensis* &  
*L. maxicana*



# Mucocutaneous Leishmaniasis

**mucocutaneous** forms of leishmaniasis , lesions can lead to partial or total destruction of the mucosa membranes of the nose, mouth and throat cavities and surrounding tissues.

Nasal stuffiness, runny nose , bleeding of nose, rectum &vagina  
Ulcer & erosion of mouth, nose, rectum, lips, gums, vaginal

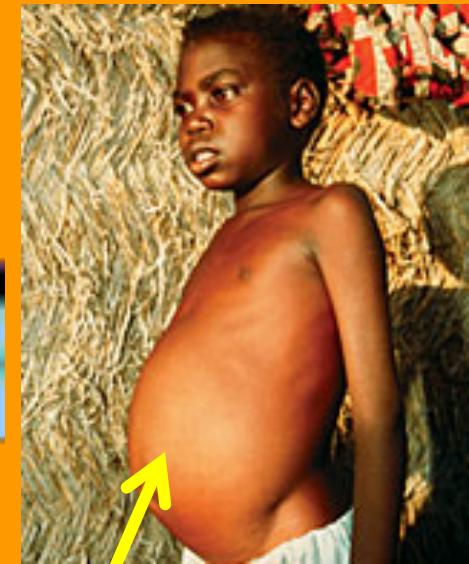


# **Visceral Leishmaniasis**

**Visceral disease (Kala-azar)**

# Visceral disease (Kala-azar)

Most severe form of disease, the disease typically starts with irregular bouts of fever, chills, and general anemia



Since leishmaniasis is primarily a disease of the reticulo-endothelial system, replacement of infected cells produces hyperplasia and consequent enlargement of the visceral organs associated with the system (e.g., spleen and liver).

*Hepatosplenomegaly*

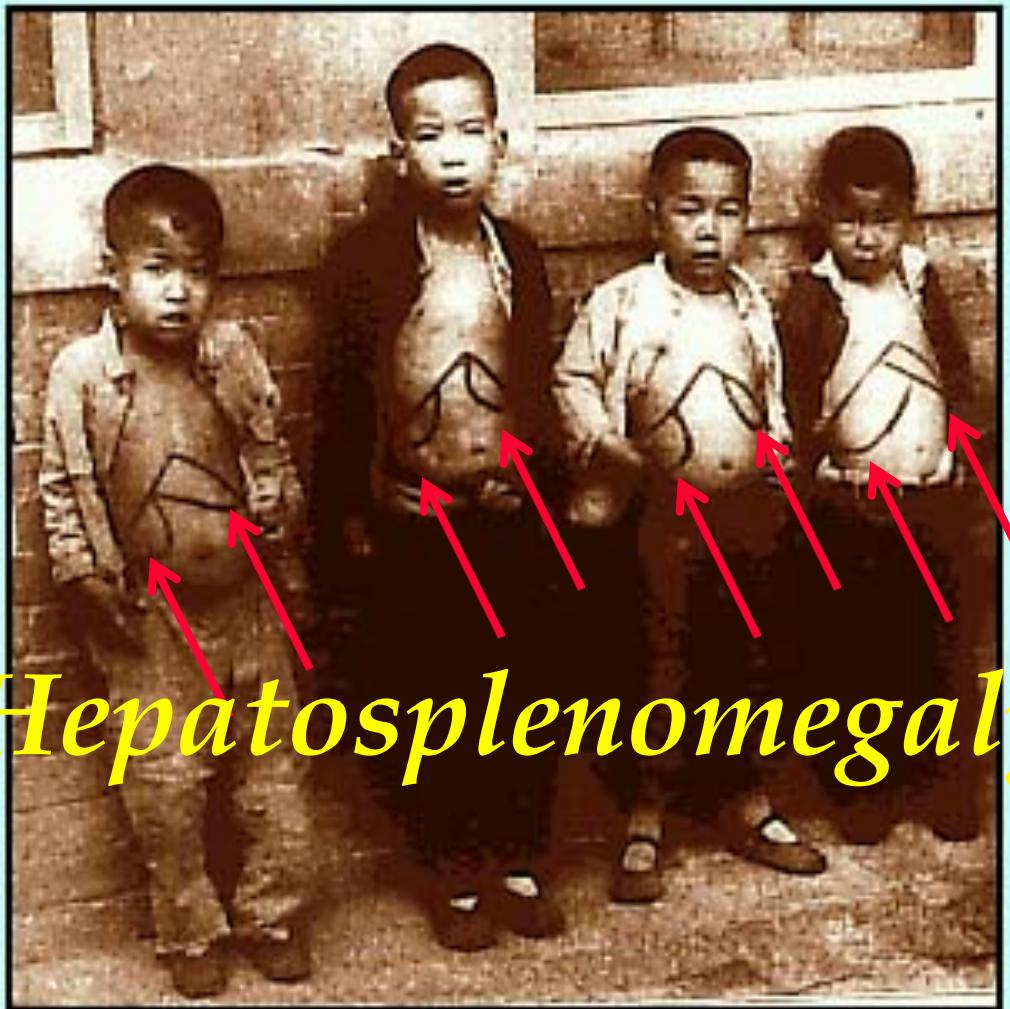
# **Post Kala Azar Dermal Leishmanoid**

Normally develops <2 years after recovery

Restricted to skin, rare but varies  
geographically

- Some people recover spontaneously
- Some people who were treated later develop Post-Kala- azar dermal leishmanoid

黑热病人照片



黑热病主要的临床表现为长期不规则发热、肝脾肿大、全血贫血。照片中患者肝、脾肿大。

皮肤型黑热病人照片



我国常见的皮肤型黑热病为结节型，皮肤结节多呈黄豆或绿豆大，结节的皮肤发红，多见于面部和颈部。



Dogs can act as reservoirs  
of *Leishmania* parasites.

They also exhibit  
symptoms of infection.

# Diagnosis

**Diagnosing Leishmaniasis can be difficult Sometimes the Lab tests are negative even if a person has Leishmaniasis.**



The  
End

- 1) Mention the stages of the life cycle of *Neglaria* sp.
- 2) Mention the habitat of *G. lamblia*
- 3) Mention the name of the disease caused by *Acanthamoeba* sp.